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Fungi on higher plants of the upper limit of the alpine zone: new species from Tian Shan

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Abstract — Fifty-two taxa of fungi were noted at the upper limit of closed vegetation in Zailijskiy Alatau Mts. (Tian Shan) in Kazakhstan. Of them three new species are described: *Protoventuria juniperina*, *Trichometasphaeria barriae*, and *Veronaea thylacospermi*. A new combination, *Didymosphaerella spartii,* with a possible *Sclerostagonospora* anamorph is proposed.

Key words — parasite, saprotroph, distribution, taxonomy

Introduction

A study of fungal diversity at the upper alpine plant limit was begun in Kazakhstan in 2005. Among the 52 taxa of fungi noted at the upper limit of closed vegetation in Tian Shan, three new species have already been described: *Cyathicula brunneospora* and *Pirottaea atrofusca* (Chlebická & Chlebicki 2007) and *Microbotryum adenopetalae* (Lutz et al. 2008). Below I propose three additional species — *Protoventuria juniperina, Trichometasphaeria barriae, Veronaea thylacospermi* — and one new combination, *Didymosphaerella spartii,* with a possible *Sclerostagonospora* anamorph.

Materials and methods

Study area: The terminal glacier foreland of the Issyk valley in Zailijskij Alatau Mts. (Tian Shan) near Almaty in southern Kazakhstan was investigated. The study was conducted on the slope of a marginal moraine (inactive ground ca 300 m before the ice margin) of the uppermost small basin at the glacier front at 3436 m elev., N43°07'52.5" E77°30'25". A distinct limit of closed vegetation was present. All native plant habitats comprised initial soil partially covered by granite rocks of various sizes (1 cm–1 m diam).

Methods: Dried material was examined under a zoom stereo microscope (Nikon SMZ 1500), and also with a light microscope Labophot 2 (Nikon) and Olympus BX-51,

at magnifications of 1000× and 2000×, and in some cases using Nomarski contrast (DIC). Microscopical observations and measurements of freehand longitudinal ascocarp sections were made in water, 3% KOH, or Lugol's solution (IKI: 1% iodine, 3% KI in water). Gelatinous sheaths of free ascospores were observed in India ink. Materials are deposited at the W. Szafer Institute of Botany of Polish Academy of Sciences in Kraków (Poland).

Species

Didymosphaerella spartii (Fabre) Chleb., **comb. nov.** FIGURES 1 A–B

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BASIONYM: *Didymosphaeria spartii* Fabre, Ann. Sci. Nat., Bot., sér. 6, 9: 83, 1879.

≡*Sphaeria spartii* Castagne, Cat. Pl. Marseille: 169, 1845, nom illegit., non Nees : Fr. 1823.

≡*Microthelia spartii* (Fabre) Kuntze, Revis Gen. Pl. 3(2): 498, 1898.

≡*Montagnula spartii* (Fabre) Aptroot, Nova Hedwigia 60: 342, 1995.

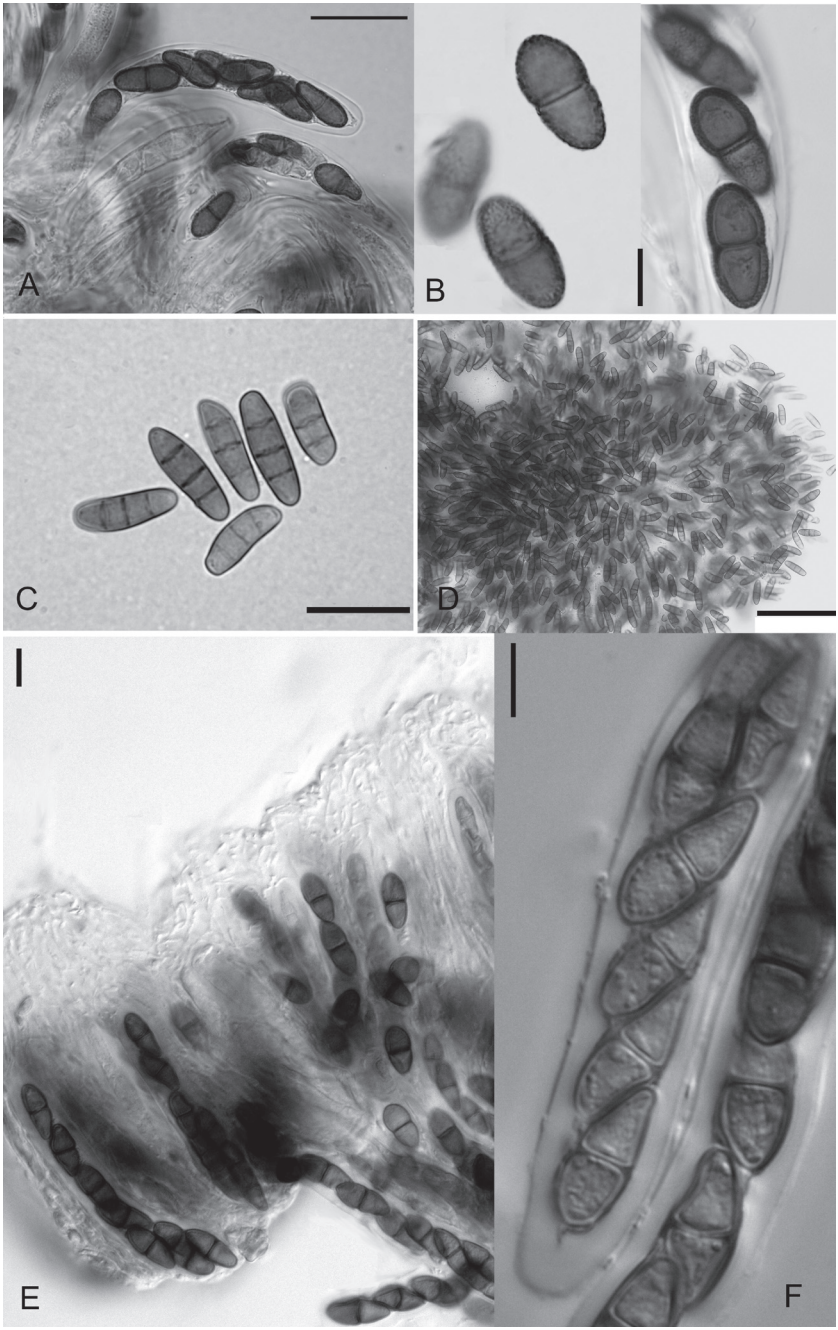
=*Didymosphaeria elbursensis* Petr., Ann. Naturh. Mus. Wien. 50: 429, 1940.

ASCOMATA globose, ca 240 µm diam., peridium wall widest in lower part, textura angulata, ASCI bitunicate, clavate 110–130 × 20–21 µm (FIG. 1A), ASCOSPORES 1-septate, slightly constricted at the septum, clear reddish brown, hemispores unequal, upper hemispore wider and slightly pointed, (20–)24–27 × 11–12 µm, wall thick and finely verruculose (FIG. 1B, left side), gelatinous sheath 3–7µm thick, uniseriate in the lower part and biseriate in upper part of ascus, INTERASCAL HYPHAE (cellular pseudoparaphyses) narrow, 1–1.4 µm diam. in its lower part and 2.2 µm diam. in upper part.

SPECIMENS EXAMINED: Kazakhstan, Tian Shan: Zailijskiy Alatau Mts., valley of Issyk (Yssyk) river, at the moraine , N43°07'52.5", E77° 30'25", 3436 m elev., 3 August 2005, on stems of *Carex griffithii* and *Anthoxanthum alpinum,* coll.: A. Chlebicki, KRAM "F" 46581.

COMMENTS — Barr (2001) lectotypified the genus *Didymosphaerella* Cooke by *Didymosphaerella longipes* (Trab.) Cooke. Placing the genus in her new family *Montagnulaceae* M.E. Barr, she transferred *Montagnula* Berl. species with two celled ascospores to *Didymosphaerella*. The Tian Shan fungus is identical to *Didymosphaeria elbursensis* (FIG.1B right side) noted on *Festuca sulcata* in Mt. Damawed in Elburs Mts. (Iran). Aptroot (1995a,b) synonymized *D. elbursensis* with *Montagnula spartii,* including also species that he considered morphologically similar occurring on palm leaves, brooms, *Ephedraceae,* and *Poaceae*. Aptroot (1995a) pointed out that ascospores in *M. spartii* have thicker walls than those in *M. opulenta* (De Not.) Aptroot [≡ *D. opulenta* (De Not.) Checa & M.E. Barr, which Barr (2001) restricted to collections from *Opuntia*].

FIG. 1. *Didymosphaerella spartii:* A. asci; B. ascospores from Tian Shan specimen (left) and Elburs Mts. specimen (right). *Sclerostagonospora* sp.: C, D. conidia. *Protoventuria juniperina:* E. asci; F. ascospores. Scale bars: A= 30 µm, C = 15 µm , E = 60 µm, other = 10 µm


A
B
C
D
E
F

Both the Tian Shan and Elburs *D. spartii* specimens possess thick walled ascospores (Fig.1b), but the ascospore size and shape differ from other taxa referred to *M spartii* by Aptroot (1995a). Aptroot (1995a) mentioned as host plants some other grasses such as *Festuca brachyphylla*, *Puccinellia angustata,* and *Stipa himalaica*. *Didymosphaerella spartii* has been noted in North America, Greenland and Asia.

Sclerostagonospora sp. Figures 1 c–d

Conidiomata immersed, Conidia pale brown with surrounded tips, three septate, 14–19 × 4–5 µm (Fig. 1 c,d).

Specimens examined: Kazakhstan, Tian Shan: Zailijskiy Alatau Mts., valley of Issyk (Yssyk) river, at the moraine , N43°07'52.5", E77° 30'25", 3436 m elev., 3 August 2005, on stems of *Festuca coelestis; Anthoxanthum alpinum*, coll.: A. Chlebicki, KRAM "F".

Comments — The close occurrence of this *Sclerostagonospora* species with the Tian Shan fungus suggests it as a possible *Didymosphaerella spartii* anamorph.

Protoventuria juniperina Chleb., **sp. nov.** Figures 1 e– f

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Mycelium superficiarum vel epidermide tectum, melanostictum, setae myceliales nullae. Ascomata 120–200 µm crassa, globosa, setae atrobrunneae 70–100 µm longae, 3 µm crassae. Asci bitunicati, clavati 70–76 × 13–14 µm, ascosporae olivaceae vel viride-olivaceae, 13–20 × 7–8 µm, 1-septatae, ad septum constrictae, exosporio laevi. Pseudoparaphyses septatae, ramosae, 1 µm crassae, ascos superantes.

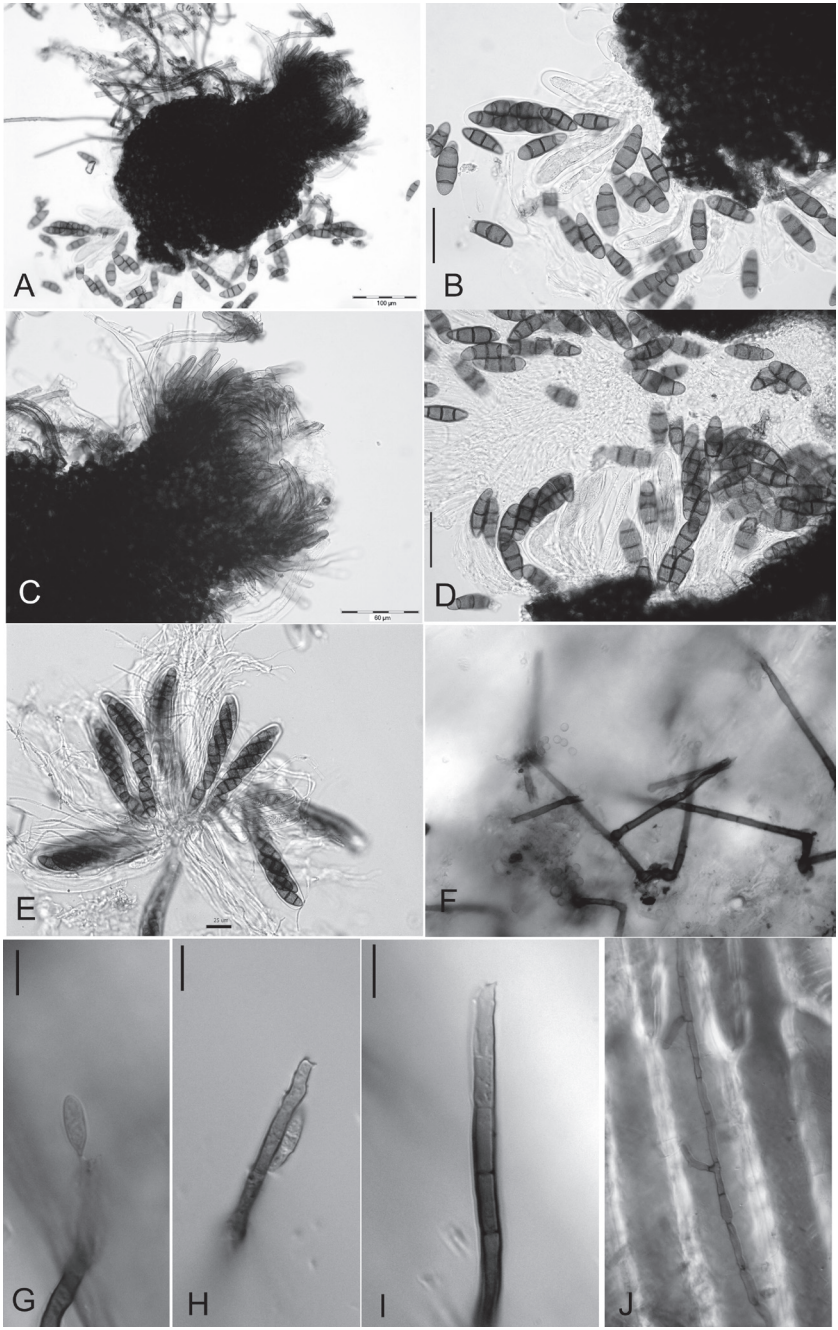
Type: Kazakhstan, Tian Shan: Zailijskiy Alatau Mts., valley of Issyk river, at the moraine, N43°07'52" E77°30'25", 3436 m elev., on dead leaves of *Juniperus sibirica*, 3 Aug. 2005, coll.: A. Chlebicki, **Holotype**-KRAM "F" 46550.

Etymology — The specific epithet refers to the host plant.

Mycelium superficial, producing subcuticular hyphae, sometimes forming dark blotches. Ascomata globose, 120–200 µm diam., surface strongly setose, Setae 70–100 µm long, 3 µm wide at the base, pointed, Asci bitunicate, clavate 70–76 × 13–14 µm, Ascospores olivaceous to greenish, septate, upper hemispore slightly wider, smooth, 13–20 × 7–8 µm, constricted at the septum, contents finely guttulate (Fig.1f). Hamathecium: pseudoparaphyses sparse, septate and branched, ca 1 µm wide, exceeding asci (Fig.1e).

Comments — *Protoventuria juniperina* somewhat resembles species in the *Herpotrichiellaceae,* which, however, possess poorly developed interascal elements or lack tissues (Barr 1972, Untereiner et al. 1995). The fungus from Tian

Fig. 2. *Trichometasphaeria barriae*: A. ascoma; B. ascospores, C. ostiole with setae, D. hamathecium and ascospores, E. asci. *Veronaea thylacospermi*: F. conidiophores with inflated basal cell, G. conidium, H, I. rachis with scattered, denticle-like coniodiogenous loci, J. mycelial hypha.
Scale bars: A = 100 µm; C = 60 µm; B, D = 40 µm; E = 25 µm; G, H, I = 10 µm


A
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Shan is referred to *Pleosporales* rather than *Dothideales* based on the presence of a distinct hamathecium. It is similar to species from the genus *Protoventuria* Berl. & Sacc. (*Venturiaceae*), which possess hypostroma, intramatrical, and subcuticular hyphae. However the *Gibbera* Fr. species previously referred to subgenus *Venturioides* M.E. Barr that Barr (1989) transferred to *Protoventuria* have some superficial and intramatrical hyphae. Holm & Holm (1977) noted two venturiaceous fungi on *Juniperus communis* leaves: *Gibbera* sp. and *Seynesiella juniperi* (Desm.) G. Arnaud. *Gibbera* sp., which most closely resembles the Tian Shan fungus, differs by producing light greenish, fusiform ascospores.

Trichometasphaeria barriae Chleb., **sp. nov.** FIGURES 2A–E

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Ascomata ex parte inclusa in substratum vel superficialia, gregaria, sphaeroidea 340–420 µm crassa, setae fuscae, apicibus pallidulis, septatae, curvatae, 70 µm longae. Peridium 24–44 µm crassum, textura angulare, asci clavati 120–140(–150) × 24–26(–27) µm, ascosporae 42–44 × 10–14(–15) µm, uniseriatae, in parte superiore biseriatae, ellipsoideae, asymmetricae, apice obtusae, 3-septatae ad septum modice constrictae, brunneolae, extremis pallidulis, exosporio laevi. Pseudoparaphyses 2 µm latae, septatae, ramosae.

TYPE: Kazakhstan, Tian Shan: Zailijskiy Alatau Mts., valley of Issyk river, at the moraine, N43°07'52" E77°30'25", 3436 m elev., on tips of stems and leaves of *Waldheimia tridactylites*, 3 Aug. 2005, coll.: A. Chlebicki, **Holotype**-KRAM "F" 46551, Isotype KRAM "F" 46552.

ETYMOLOGY — the epithet refers to the late Margaret E. Barr.

ASCOMATA (FIG. 2A) partially embedded in the substratum, rarely erumpent, gregarious, 340–420 µm wide, ca 500 µm high, wall 24–44 µm thick, composed of an external dark layer with 3 rows of cells and an internal light layer with 2–3 rows of cells, textura angularis, subiculum absent, with a distinct OSTIOLE ca 160 µm diam, 90–100 µm high, covered by short (up to 70 µm long and 4–6 µm wide) straight or slightly curved septate setae (FIG. 2C), paler at the tips and sometimes slightly rough, surface of ascomata covered by very long, curved, darker, septate, thick walled and downward growing hyphae, 5–6 µm diam. at the base, ASCI clavate, 120–140(–150) × 24–26(–27) µm (FIG. 2E), ASCOSPORES 37–44(–45) × 10–14(–15) µm, 3-septate, constricted at the supramedian septum, slightly asymmetric, pale brown, distal cells slightly paler (FIG. 2B) PSEUDOPARAPHYSES ca 2 µm diam., branched and septate, very abundant in the centrum, hyaline, their cells inside granulate (FIG. 2D).

COMMENTS — Yuan & Barr (1994) described a new species, *T. papillisetosa* Z.Q. Yuan & M.E. Barr, also from Tian Shan (China) but on decorticated branches of *Pentaphylloides fruticosa*. Both *T. barriae* and *T. papillisetosa* produce similar ascomata with septate, erect setae with paler tips and larger ascospores. The smooth-walled ascospores help distinguish *T. barriae* from *T. papillisetosa,* which has verrucose ascospores.

Veronaea thylacospermi Chleb., **sp. nov.** FIGURES 2F–J

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Coloniae hypophyllae, effusae, brunneae. Conidiophora singularia, erecta, recta vel flexuosa, 1–3(–8) septata, pallide brunnea, usque 35–100 µm longa, 3–4 µm crassa, sub conidiophoris ad 6–10 µm, apicem versus pallidiora. Cellulae conidiogenae 0.3 µm longae et 0.5 µm latae. Conidia obclavata, subhyalina, verruculosa, 1–septata, basi obconico-truncata, apicem versus rotundata 12–14 × 3–4 µm.

TYPE: Kazakhstan, Tian Shan: Zailijskiy Alatau Mts., valley of Issyk river, at the moraine, N43°07'52" E77°30'25", 3436 m elev., on leaves of *Thylacospermum caespitosum*, 3 Aug. 2005, coll.: A. Chlebicki, **Holotype**-KRAM "F" 46601.

ETYMOLOGY — Refers to the host plant, *Thylacospermum caespitosum*.

CONIDIOPHORES simple, smooth, brown, 1–3(–8) septate, 35–100 × 3–4 µm, basal cell inflated 6–10 µm wide (FIG. 2F), fertile part taller than basal part, forming slightly flexuose rachis with scattered, hyaline and small. Apically pointed denticle-like CONIDIOGENOUS LOCI, 0.3 µm high, 0.5 µm wide (FIG. 2H,I). CONIDIA hyaline 12–14 × 3–4 µm, two celled, lower cell longer and wider than upper one, wall slightly verruculose (FIG. 2G). Paler mycelial hyphae distributed inside host cells (FIG. 2J).

COMMENTS — No *Veronaea* species have been previously reported from *T. caespitosum*. The conidia of *V. thylacospermi* are similar to those of *V. caricis* M.B. Ellis, illustrated by Ellis (1976). This fungus is a transitional form between the genera *Veronaea* and *Myrmecridium* (Arzanlou et al. 2007); however its two celled conidia devoid of gelatinous sheath indicate a relationship with *Veronaea*.

Acknowledgments

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